

Amendments to the Claims:

This listing of claims will replace all prior versions, and listings, of claims in the application:

Listing of Claims:

1. (Currently amended) An isolated and purified microbial organism, wherein said microbial organism is capable of fermenting malic acid to lactic acid,

and wherein said microbial organism when placed in a medium containing a predetermined amount of citric acid in the range of 1 to 5000 mg/L, is only capable of degrading at the most 80% of said citric acid,

and wherein the microbial organism ~~has at least one of the following characteristics, is capable, under suitable growth conditions, of adaption to,~~ when said microbial organism in a frozen or freeze dried state is added directly into a fermented fruit juice:

- i) a survival rate which is at least 1% after two days at 23°C in a fermented sterile fruit juice with a pH of less than 4 and comprising at least 12 vol% ethanol, and/or
- ii) a survival rate which is at least 70% after two days at 17°C in a fermented sterile fruit juice with a pH of less than 4 comprising at least 13.9 vol% ethanol,

and wherein the microbial organism is selected from species belonging to the group of genera consisting of *Lactobacillus*, *Pediococcus*, and *Oenococcus*.

2. (Original) The microbial organism according to claim 1, wherein said microbial organism is an *Oenococcus oeni* strain.
3. (Cancelled)
4. (Currently amended) The microbial organism according to claim 1, wherein the ~~characteristic is~~ organism has a survival rate which is at least 10% after two days at 23°C in a wine prepared by yeasting a sterile grape fruit juice without added sulphite, said wine having an ethanol content of 12.0 vol%, pH 3.4, below 5 g/L residual sugar, 3.3 g/L of malic acid, and 450 mg/L of citric acid.
5. (Cancelled)
6. (Currently amended) The microbial organism according to claim 1, wherein the ~~characteristic is~~ organism has a survival rate which is in the range of 70% to 100% after two days at 18°C in a wine prepared with 30 ppm SO<sub>2</sub> added before the alcoholic fermentation, said wine having an ethanol content of 13.8 vol%, pH 3.5, 1.3 g/L malic acid, and 340 mg/L of citric acid.
7. (Currently amended) The microbial organism according to claim 1, wherein the ~~characteristic is~~ organism has a survival rate which is at least 80% after two days at 17°C in a wine prepared without SO<sub>2</sub> added, said wine having an ethanol content of 13.9 vol%, pH 3.6, 1.7 g/L malic acid, and 320 mg/L of citric acid.

8. (Currently amended) The microbial organism according to claim 1, wherein said microbial organism when placed in a liquid composition comprising a predetermined amount of malic acid in the range of 1-50 mg/L, is capable of degrading at least 90% of said malic acid.
9. (Original) The microbial organism according to claim 1, wherein said microbial organism is only capable of degrading at the most 50% of said citric acid.
10. (Cancelled)
11. (Original) The microbial organism according to claim 1, wherein said microbial organism reduces the citric acid content by less than 50% within 50 days, when added directly in a frozen or freeze dried state to a fermented fruit juice at a concentration of CFUs in the range of  $1 \times 10^6$  to  $5 \times 10^7$  per ml, wherein said fermented fruit juice is prepared by yeasting a sterile fruit juice without added sulphite resulting in a fermented fruit juice having an ethanol content of 12.0 vol%, pH 3.4, below 5 g/L residual sugar, 3.3 g/L of malic acid, and 450 mg/L of citric acid.
12. (Cancelled)
13. (Cancelled)
14. (Cancelled)
15. (Previously presented) The microbial organism according to claim 1, wherein said organism is selected from the group

consisting of strains deposited under the accession numbers DSM 15569, DSM 15570, and DSM 15571.

16. (Withdrawn) A method of preferentially degrading malic acid over citric acid in a liquid composition comprising malic acid and citric acid, said method comprising the steps of

- i) Providing a liquid composition comprising malic acid and citric acid;
- ii) Providing a microbial organism according to claim 1, wherein said microbial organism has been frozen or freeze dried,
- iii) Adding said freeze dried or frozen microbial organism directly to said liquid composition
- iv) incubating said liquid composition and said microbial organism under conditions which allow degradation of at least 70% of the malic acid,
- v) thereby obtaining a final liquid composition comprising less than 30% of the initial malic acid and at least 20% of the initial citric acid.

17. (Cancelled)

18. (Withdrawn) The method according to claim 16, wherein the liquid composition is grape juice or fermented grape juice.

19. (Withdrawn) The method according to claim 16, wherein the liquid composition has a pH in the range of 2 to 5.

20. (Withdrawn) The method according to claim 16, wherein the liquid composition comprises in the range of 5 to 15 vol% ethanol.

21. (Cancelled)

22. (Withdrawn) The method according to claim 16, wherein the liquid composition comprises in the range of 1 to 10 g/L malic acid.

23. (Withdrawn) The method according to claim 16, wherein the liquid composition comprises in the range of 50 to 2000 mg/L citric acid.

24. (Withdrawn) The method according to claim 16, wherein the final liquid composition comprises at least 50% of the initial citric acid.

25. (Withdrawn) The method according to claim 16, wherein the final liquid composition comprises less than 20% of the initial malic acid.

26. (Withdrawn) The method according to claim 16, wherein the microbial organism is added at a concentration of less than  $5 \times 10^7$  CFU per ml of the liquid composition.

27. (Cancelled)

28. (Cancelled)

29. (Withdrawn) The method according to claim 18, wherein the fermented grape juice is selected from the group consisting of red wines, white wines and sparkling wines.

30. (Cancelled)

31. (Withdrawn - Currently Amended) The method according to claim 30 16, wherein step iv) comprises incubation for a longer period of time than required for completion of malolactic fermentation.

32. (Withdrawn) A method of inducing malolactic fermentation during wine production, comprising the steps of

- i) Providing a grape juice or a fermented grape juice
- ii) Providing a microbial organism according to claim 1,
- iii) Incubating said grape juice or fermented grape juice with said microbial organism under conditions which allow degradation of malic acid,
- iv) thereby inducing malolactic fermentation.

33. (Cancelled)

34. (Withdrawn - Currently Amended) The method according to claim 33 16, wherein said microbial organism in a frozen or freeze-dried state is added directly to said grape juice or a fermented grape juice.

35. (Withdrawn) The method according to claim 32, wherein the microbial organism is added at a concentration of less than  $5 \times 10^7$  CFU per ml of the grape juice or a fermented grape juice.

36. (Cancelled)

37. (Withdrawn - Currently amended) The method according to claim 32, wherein the grape juice or a fermented grape juice has a pH in the range of 2 to 5[[],].

38. (Withdrawn - Currently amended) The method according to claim 32, wherein the grape juice or a fermented grape juice comprises in the range of 5 to 15 vol% ethanol[[],].
39. (Cancelled)
40. (Withdrawn) The method according to claim 32, wherein the grape juice or a fermented grape juice comprises in the range of 1 to 10 g/L malic acid.
41. (Withdrawn) The method according to claim 32, wherein the grape juice or a fermented grape juice comprises in the range of 50 to 2000 mg/L citric acid.
42. (Withdrawn) The method according to claim 32, wherein the wine is selected from the group consisting of red wines, white wines and sparkling wines.
43. (Previously presented) A concentrate of microbial organisms comprising the microbial organism according to claim 1, wherein said concentrate has a content of colony forming units being in the range of  $10^9$  to  $10^{12}$  per g.
44. (Cancelled)
45. (Previously presented) The concentrate according to claim 43, wherein said microbial organism has been propagated in an adaptation medium comprising at least 6% sugar.
46. (Cancelled)
47. (Cancelled)

48. (Previously presented) The concentrate according to claim 45, wherein said adaptation medium comprises at least 3% glucose and at least 3% fructose.
49. (Previously presented) The concentrate according to claim 45, wherein said microbial organism has been propagated in said adaptation medium for at least 12 hours.
50. (Withdrawn) A method of producing a microbial organism according to claim 1, wherein said method comprises the steps of
- i) Providing a microbial organism resistant to a pH below 5 and an ethanol concentration of at least 8%,
  - ii) Subjecting said microbial organism to mutagenesis, thereby obtaining more than one different mutated microbial organism
  - iii) Selecting mutated microbial organisms capable of fermenting malic acid to lactic acid, wherein said microbial organism when placed in a medium containing a predetermined amount of citric acid is only capable of degrading at the most 80% of said citric acid, and wherein the microbial organism has at least one of the following characteristics, when said microbial organism in a frozen or freeze dried state is added directly into a fermented fruit juice:
- a) a survival rate which is at least 1% after two days at 23°C in a fermented sterile fruit juice comprising at least 12 vol% ethanol;

b) a survival rate which is at least 70% after two days at 17°C in a fermented sterile fruit juice comprising at least 13.9 vol% ethanol

51. (Withdrawn) The method according to claim 50, wherein said microbial organism is resistant to pH 3.2.

52. (Withdrawn) The method according to claim 50, wherein said microbial organism is resistant to an ethanol concentration of 13 vol%.

53. (Cancelled)

54. (Cancelled)

55. (Withdrawn) A method of preparing a dried microbial organism capable of fermenting malic acid to lactic acid, which has reduced citric acid degrading activity and which is capable of survival after direct inoculation into fermented fruit juice, said method comprising the steps of

- i) Providing a microbial organism according to claim 1,
- ii) Providing an adaptation medium comprising at least 6% sugar
- iii) Propagating said microbial organism in said adaptation medium under conditions allowing growth of said microbial organism
- iv) Harvesting said microbial organism
- v) drying said microbial organism.

56. (Cancelled)

57. (Cancelled)

58. (Withdrawn) The method according to claim 55, wherein said adaptation medium comprises at least 3% glucose and at least 3% fructose.

59. (Withdrawn) The method according to claim 55, wherein said microbial organism is propagated in said adaptation medium for at least 12 hours.

60. (Cancelled)

61. (Cancelled)

62. (Cancelled)

63. (Withdrawn) An activation solution comprising  
i) A nitrogen source  
ii) In the range of 60 to 140 g sugar per L  
iii) In the range of  $5 \times 10^8$  and  $5 \times 10^{10}$  colony forming units per ml of a microbial organism capable of fermenting at least one fermentable compound  
iv) a chemical compound with buffering capacity, wherein the solution has a pH in the range of 4 to 6.

64. (Withdrawn) The activation solution according to claim 63, wherein said fermentable compound is malic acid.

65. (Withdrawn) The activation solution according to claim 63, wherein said microbial organism is capable of fermenting malic acid to lactic acid.

66. (Withdrawn) The activation solution according to claim 63, wherein said microbial organism is selected from the group consisting of bacteria belonging to the *Oenococcus* family and the *Lactobacillus* family.
67. (Cancelled)
68. (Withdrawn) The activation solution according to claim 63, wherein the microbial organism is selected from the group consisting of DSM 15568, DSM 7008, DSM 15569, DSM 15570, and DSM 15571.
69. (Withdrawn) The activation solution according to claim 63, wherein said microbial organism is selected from the group consisting of MBR Alpha, MBR Beta, MBR 31, MBR 41, MBR OSU, Inobacter (IB), OSU, ProVino, MCW, 3X, MT01, Viniflora oenos and Viniflora CH35.
70. (Withdrawn) The activation solution according to claim 63, wherein the sugar is selected from the group consisting of fructose and glucose.
71. (Withdrawn - Currently amended) The activation solution according to claim 63, wherein the glucose content of the solution is in the range of 30 g to 100 g glucose per liter solution.
72. (Withdrawn) The activation solution according to claim 63, wherein the fructose content of the solution is in the range of 30 g to 100 g fructose per liter solution.

73. (Withdrawn) The activation solution according to claim 63, wherein said chemical compound is selected from the group consisting of tartaric acid, malic acid, lactic acid, phosphate and citrate.

74. (Withdrawn - Currently amended) A dry activation composition, wherein in the range of 80 to 200 g of said dry activation composition comprises

- i) a nitrogen source
- ii) in the range of 60 to 140 g sugar
- iii) in the range of  $5 \times 10^{11}$  and  $5 \times 10^{13}$  colony forming units of a microbial organism capable of fermenting a fermentable compound,
- iv) a chemical ~~compoound~~ compound with buffering capacity, wherein the chemical compound is capable of buffering a solution to a pH in the range of 4.0 to 6.0.

wherein addition of 1 L water to said dry activation compositions results in an activation solution according to claim 63.

75. (Withdrawn) The composition according to claim 74, wherein in the range of 110 to 150 g of said dry activation composition comprises in the range of 80 to 110 g sugar.

76. (Withdrawn) The composition according to claim 74, wherein the sugar is selected from the group consisting of fructose and glucose.

77. (Cancelled)

78. (Cancelled)

79. (Withdrawn) The composition according to claim 74, wherein the glucose concentration is 40 to 80 g/L and the fructose concentration is 40 to 80 g/L.
80. (Withdrawn) The composition according to claim 74, wherein the fermentable compound is malic acid.
81. (Withdrawn) The composition according to claim 74, wherein said microbial organism when incubated in a liquid composition comprising a predetermined amount of malic acid is capable of fermenting at least 50% of said malic acid.
82. (Withdrawn) The composition according to claim 74, wherein said microbial organism is selected from the group consisting of bacteria belonging to the *Oenococcus* family and the *Lactobacillus* family.
83. (Cancelled)
84. (Withdrawn) The composition according to claim 74, wherein the microbial organism is selected from the group consisting of DSM 15568, DSM 7008, DSM 15569, DSM 15570, and DSM 15571.
85. (Withdrawn) The composition according to claim 74, wherein the microbial organism is selected from the group consisting of MBR Alpha, MBR Beta, MBR 31, MBR 41, MBR OSU, Inobacter (IB), OSU, ProVino, MCW, 3X, MT01, Viniflora oenos and Viniflora CH35.
86. (Withdrawn) The composition according to claim 74, wherein the microbial organism after activation for more than 5 hours in the activation solution has a survival rate which

is at least 3% after two days at 23°C when inoculated into a fermented fruit juice having an ethanol content of at least 12.0 vol%.

87. (Withdrawn) The composition according to claim 86, wherein activation is in the range of 8 to 48 hours.

88. (Withdrawn) The composition according to claim 86, wherein activation is performed at a temperature in the range of 18°C to 25°C.

89. (Cancelled)

90. (Withdrawn) The composition according to claim 86, wherein the survival rate is at least 33% after two days at 23°C when inoculated into a fermented fruit juice prepared by yeasting a sterile grape fruit juice without added sulphite, said fermented fruit juice having an ethanol content of 12.0 vol%, pH 3.4, below 5 g/L residual sugar, 3.3 g/L of malic acid, and 450 mg/L of citric acid.

91. (Withdrawn) The composition according to claim 86, wherein the survival rate is at least 94% after two days at 23°C when inoculated into a fermented fruit juice prepared by yeasting a sterile grape fruit juice without added sulphite, said fermented fruit juice having an ethanol content of 12.0 vol%, pH 3.4, below 5 g/L residual sugar, 3.3 g/L of malic acid, and 450 mg/L of citric acid.

92. (Withdrawn) The composition according to claim 74, wherein said chemical compound is selected from the group consisting

of tartaric acid, malic acid, lactic acid, phosphate and citrate.

93. (Withdrawn) The composition according to claim 74, wherein said composition furthermore comprises a salt.

94. (Withdrawn) A method of inducing fermentation in a liquid composition comprising a fermentable compound comprising the steps of

- i) Providing a dry composition according to claim 74, wherein said microbial organism is capable of fermenting said fermentable compound
- ii) Adding water to said dry composition, thereby obtaining an activation solution
- iii) Incubating said activation solution for an activation time under activation conditions
- iv) Providing a liquid composition comprising said fermentable compound
- v) Inoculating said liquid composition with said activation solution
- vi) Thereby inducing fermentation in said liquid composition.

95. (Withdrawn) The method according to claim 94, wherein the liquid composition is a fruit juice or a fermented fruit juice.

96. (Cancelled)

97. (Cancelled)

98. (Withdrawn) The method according to claim 94, wherein the activation time is in the range of 8 to 48 hours.

99. (Withdrawn - Currently amended) The method according to claim 94, wherein activation conditions comprises incubation at a temperature in the range of 10°C to 40°C[[],].

100. (withdrawn) The method according to claim 94, wherein the fermentable compound is malic acid.

101. (New) The organism of claim 1, wherein said growth conditions suitable to adaptation to the survival rate of (i) and/or (ii) comprise propagation of said organism in an adaptation medium comprising at least 6% sugar.

102. (New) The organism of claim 101, wherein said adaptation medium comprises at least 3% glucose and at least 3% fructose.

103. (New) The organism of claim 101, wherein the organism is propagated in said adaptation medium for at least 12 hours.